F "ENT COOPERATION TREA

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and Administrative Instructions, Section 422)

To:

BIRD, William, E.
Bird Goën & Co
Vilvoordsebaan 92
B-3020 Winksele
BELGIQUE

Date of mailing (day/month/year)
07 June 2001 (07.06.01)

Applicant's or agent's file reference
T1639-PCT

IMPORTANT NOTIFICATION

International application No.
PCT/BE00/00120

International filing date (day/month/year)
06 October 2000 (06.10.00)

1. The following indications appeared on record concerning:

[X] the applicant [ ] the inventor [ ] the agent [ ] the common representative

Name and Address
TIGENIX N.V.
Velpestraat 17
B-3360 Bierbeek
Belgium

State of Nationality: BE
State of Residence: BE
Telephone No.:
Facsimile No.:
Teleprinter No.:

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

[ ] the person [ ] the name [X] the address [ ] the nationality [ ] the residence

Name and Address
TIGENIX N.V.
Boetsenberg 18
B-3053 Haasrode
Belgium

State of Nationality: BE
State of Residence: BE
Telephone No.:
Facsimile No.:
Teleprinter No.:

3. Further observations, if necessary:

4. A copy of this notification has been sent to:

[X] the receiving Office
[ ] the International Searching Authority
[ ] the International Preliminary Examining Authority
[X] the designated Offices concerned
[ ] the elected Offices concerned
[ ] other:

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No.: (41-22) 740.14.35

Authorized officer
R. Chrem

Telephone No.: (41-22) 338.83.38

Copy for the Elected Office (EO/US) PCT/BE00/00120

I :ENT COOPERATION TREA :

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and Administrative Instructions, Section 422)

To:

BIRD, William, E.
Bird Goën & Co
Vilvoordsebaan 92
B-3020 Winksele
BELGIQUE

Date of mailing (day/month/year)
23 April 2002 (23.04.02)

Applicant's or agent's file reference
T1639-PCT

IMPORTANT NOTIFICATION

International application No.
PCT/BE00/00120

International filing date (day/month/year)
06 October 2000 (06.10.00)

1. The following indications appeared on record concerning:

[X] the applicant [X] the inventor [ ] the agent [ ] the common representative

Name and Address
DELL'ACCIO, Francesco
IJzerenmolenstraat 32/125
B-3001 Heverlee
Belgium

State of Nationality: IT
State of Residence: BE
Telephone No.:
Facsimile No.:
Teleprinter No.:

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

[ ] the person [ ] the name [ ] the address [ ] the nationality [ ] the residence

Name and Address
DELL'ACCIO, Francesco
Celestijnenlaan 3/51
B-3001 Heverlee
Belgium

State of Nationality: IT
State of Residence: BE
Telephone No.:
Facsimile No.:
Teleprinter No.:

3. Further observations, if necessary:

4. A copy of this notification has been sent to:

[X] the receiving Office
[ ] the International Searching Authority
[ ] the International Preliminary Examining Authority
[ ] the designated Offices concerned
[X] the elected Offices concerned
[ ] other:

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No.: (41-22) 740.14.35

Authorized officer
Lazar Joseph PANAKAL

Telephone No.: (41-22) 338.83.38

P. ENT COOPERATION TREA

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF ELECTION

(PCT Rule 61.2)

To:

Commissioner
US Department of Commerce
United States Patent and Trademark Office, PCT
2011 South Clark Place Room CP2/5C24
Arlington, VA 22202
ETATS-UNIS D'AMERIQUE

in its capacity as elected Office

Date of mailing (day/month/year) 26 June 2001 (26.06.01)	
International application No. PCT/BE00/00120	**Applicant's or agent's file reference** T1639-PCT
International filing date (day/month/year) 06 October 2000 (06.10.00)	**Priority date** (day/month/year) 06 October 1999 (06.10.99)
Applicant LUYTEN, Frank et al	

1. The designated Office is hereby notified of its election made:

[X] in the demand filed with the International Preliminary Examining Authority on:

13 April 2001 (13.04.01)

[ ] in a notice effecting later election filed with the International Bureau on:

2. The election [X] was

[ ] was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland**	Authorized officer Pascal Piriou
Facsimile No.: (41-22) 740.14.35	Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

PCT

INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference	FOR FURTHER ACTION	see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.
T1639-PCT		
International application No.	International filing date *(day/month/year)*	(Earliest) Priority Date *(day/month/year)*
PCT/BE 00/00120	06/10/2000	06/10/1999
Applicant		
TIGENIX N.V. et al.		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 6 sheets.

[X] It is also accompanied by a copy of each prior art document cited in this report.

1. **Basis of the report**

   a. With regard to the **language,** the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

   [ ] the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

   b. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international search was carried out on the basis of the sequence listing :

   [ ] contained in the international application in written form.

   [ ] filed together with the international application in computer readable form.

   [ ] furnished subsequently to this Authority in written form.

   [ ] furnished subsequently to this Authority in computer readble form.

   [ ] the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   [ ] the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

2. [X] **Certain claims were found unsearchable** (See Box I).

3. [ ] **Unity of invention is lacking** (see Box II).

4. With regard to the **title,**

   [X] the text is approved as submitted by the applicant.

   [ ] the text has been established by this Authority to read as follows:

5. With regard to the **abstract,**

   [X] the text is approved as submitted by the applicant.

   [ ] the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. The figure of the **drawings** to be published with the abstract is Figure No. _____

   [ ] as suggested by the applicant.

   [ ] because the applicant failed to suggest a figure.

   [ ] because this figure better characterizes the invention.

   [X] None of the figures.

INTERNATIONAL SEARCH REPORT

ational application No.
PCT/BE 00/00120

B x I Observati ns where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☒ Claims Nos.: 1-30 in part
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 1-30 in part

The expression "or a marker co-expressed and/or co-detectable with this marker" covers such a large number of possibilities that no meaningful search can be carried out. The search is restricted to "expressed morphogeneic protein" as a marker.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

International Application No
PC[redacted]E 00/00120

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12N5/06 A61K9/00 A61P19/02 A61P19/04 G01N33/53

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 A61K A61P C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, WPI Data, PAJ, BIOSIS, MEDLINE, CHEM ABS Data, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 35022 A (OSIRIS THERAPEUTICS INC) 13 August 1998 (1998-08-13) * see claims 1 and 5-7, page 4, page 16 last paragraph and example 1 * ---	1,4-6,8, 10,11, 21, 23-25, 28-30
X	WO 96 41620 A (YAYON AVNER ;NEVO ZVI (IL); UNIV RAMOT (IL); YEDA RES & DEV (IL)) 27 December 1996 (1996-12-27) * see claims 1-11, 14 and page 7 * --- -/--	1,2,4-6, 8,10-14, 16-18, 20,22-30

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
11 January 2001	19/02/01

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Gore, V

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
X	WO 97 26326 A (BOEHRINGER MANNHEIM CORP) 24 July 1997 (1997-07-24) * see cl. 48 and 54 *	1,4-6,8, 10-14, 16-18, 20-30
X	US 5 811 094 A (HAYNESWORTH STEPHEN E ET AL) 22 September 1998 (1998-09-22)	1,2,4-6, 8,10-14, 16-18, 20-30
Y	* see claims 1-2, 5-7 and 23, col.4, col.2 line 57 to col.3 line 3, col.38 line 53 to col.39 line 34, col. 44 line 66 to col. 45 line 32 *,	1-30
X	WO 96 07733 A (MACKAY VIVIAN L ;MOORE EMMA E (US); ZYMOGENETICS INC (US)) 14 March 1996 (1996-03-14) * see claims 16-19, pages 4-5 and 37-38 *	1,4-6,8, 10,11, 21,24, 25,28-30
X,P	US 5 972 703 A (LONG MICHAEL W ET AL) 26 October 1999 (1999-10-26) * see claims 48-50 and col.11-12 *	1,4-6,8, 10-14, 16-18, 20-30
X	WO 97 07200 A (UNIV LELAND STANFORD JUNIOR ;BARRES BARBARA A (US)) 27 February 1997 (1997-02-27) * claim 7 *	1,2,8, 10,11, 21-25, 28-30
X	NEVO Z ET AL: "THE MANIPULATED MESENCHYMAL CELLS IN REGENERATED SKELETAL TISSUES" CELL TRANSPORTATION,US,ELSEVIER SCIENCE, NEW YORK, vol. 7, no. 1, February 1998 (1998-02), pages 63-70, XP002112185 ISSN: 0963-6897 * see abstract and page 63 right col. *	2-6, 12-14, 16-18, 20-30
A	WO 99 28443 A (UNIV UTAH RES FOUND) 10 June 1999 (1999-06-10) * see claims 26-27 *	1-30

-/--

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
Y	ERLACHER L. ET AL.: "Cartilage-derived morphogenetic proteins and osteogenic protein-1 differentially regulate osteogenesis." JOURNAL OF BONE AND MINERAL RESEARCH., vol. 13, 1998, pages 383-392, XP000972916 * see abstract and page 383 right col. to page 384 left col. *	1-30

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 00/00120

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
WO 9835022	A	13-08-1998	AU	6144498 A	26-08-1998
WO 9641620	A	27-12-1996	AU	715996 B	17-02-2000
			AU	6014496 A	09-01-1997
			AU	721773 B	13-07-2000
			AU	6014596 A	09-01-1997
			CA	2223701 A	27-12-1996
			CA	2224229 A	27-12-1996
			CN	1195982 A	14-10-1998
			EP	0833620 A	08-04-1998
			EP	0836380 A	22-04-1998
			WO	9641523 A	27-12-1996
			JP	11508358 T	21-07-1999
			JP	11507828 T	13-07-1999
WO 9726326	A	24-07-1997	EP	0877795 A	18-11-1998
			JP	2000503542 T	28-03-2000
US 5811094	A	22-09-1998	US	5486359 A	23-01-1996
			US	5226914 A	13-07-1993
			US	5197985 A	30-03-1993
			US	6010696 A	04-01-2000
			US	6087113 A	11-07-2000
			US	5733542 A	31-03-1998
			US	5837539 A	17-11-1998
			AU	669850 B	27-06-1996
			AU	2252492 A	12-01-1993
			CA	2111845 A	23-12-1992
			EP	0592521 A	20-04-1994
			JP	7500001 T	05-01-1995
			WO	9222584 A	23-12-1992
WO 9607733	A	14-03-1996	US	5683906 A	04-11-1997
			US	5648219 A	15-07-1997
			AU	3586595 A	27-03-1996
			CA	2199377 A	14-03-1996
			EP	0804554 A	05-11-1997
			JP	10505498 T	02-06-1998
			US	5830682 A	03-11-1998
US 5972703	A	26-10-1999	AU	712247 B	04-11-1999
			AU	3244295 A	07-03-1996
			CA	2200197 A	22-02-1996
			EP	0804551 A	05-11-1997
			WO	9605290 A	22-02-1996
WO 9707200	A	27-02-1997	AU	7152296 A	12-03-1997
WO 9928443	A	10-06-1999	EP	1034255 A	13-09-2000

TRAITE DE [redacted]PERATION EN MATIERE DE B ETS

PCT

RAPPORT DE RECHERCHE INTERNATIONALE

(article 18 et règles 43 et 44 du PCT)

Référence du dossier du déposant ou du mandataire	**POUR SUITE A DONNER** voir la notification de transmission du rapport de recherche internationale (formulaire PCT/ISA/220) et, le cas échéant, le point 5 ci-après	
Demande internationale n° PCT/BE 00/ 00122	Date du dépôt international *(jour/mois/année)* 11/10/2000	(Date de priorité (la plus ancienne) *(jour/mois/année)* 13/10/1999
Déposant OST, Christian		

Le présent rapport de recherche internationale, établi par l'administration chargée de la recherche internationale, est transmis au déposant conformément à l'article 18. Une copie en est transmise au Bureau international.

Ce rapport de recherche internationale comprend 2 feuilles.

[X] Il est aussi accompagné d'une copie de chaque document relatif à l'état de la technique qui y est cité.

1. **Base du rapport**

a. En ce qui concerne la **langue,** la recherche internationale a été effectuée sur la base de la demande internationale dans la langue dans laquelle elle a été déposée, sauf indication contraire donnée sous le même point.

[ ] la recherche internationale a été effectuée sur la base d'une traduction de la demande internationale remise à l'administration.

b. En ce qui concerne **les séquences de nucléotides ou d'acides aminés** divulguées dans la demande internationale (le cas échéant), la recherche internationale a été effectuée sur la base du listage des séquences :

[ ] contenu dans la demande internationale, sous forme écrite.

[ ] déposée avec la demande internationale, sous forme déchiffrable par ordinateur.

[ ] remis ultérieurement à l'administration, sous forme écrite.

[ ] remis ultérieurement à l'administration, sous forme déchiffrable par ordinateur.

[ ] La déclaration, selon laquelle le listage des séquences présenté par écrit et fourni ultérieurement ne vas pas au-delà de la divulgation faite dans la demande telle que déposée, a été fournie.

[ ] La déclaration, selon laquelle les informations enregistrées sous forme déchiffrable par ordinateur sont identiques à celles du listage des séquences présenté par écrit, a été fournie.

2. [ ] **Il a été estimé que certaines revendications ne pouvaient pas faire l'objet d'une recherche** (voir le cadre I).

3. [ ] **Il y a absence d'unité de l'invention** (voir le cadre II).

4. En ce qui concerne le **titre,**

[X] le texte est approuvé tel qu'il a été remis par le déposant.

[ ] Le texte a été établi par l'administration et a la teneur suivante:

5. En ce qui concerne **l'abrégé,**

[X] le texte est approuvé tel qu'il a été remis par le déposant

[ ] le texte (reproduit dans le cadre III) a été établi par l'administration conformément à la règle 38.2b). Le déposant peut présenter des observations à l'administration dans un délai d'un mois à compter de la date d'expédition du présent rapport de recherche internationale.

6. La figure **des dessins** à publier avec l'abrégé est la Figure n° 1

[X] suggérée par le déposant.

[ ] parce que le déposant n'a pas suggéré de figure.

[ ] parce que cette figure caractérise mieux l'invention.

[ ] Aucune des figures n'est à publier.

RAPPORT DE RECHERCHE INTERNATIONALE

Demande internationale No
BE 00/00122

A. CLASSEMENT DE L'OBJET DE LA DEMANDE
CIB 7 F24F7/06

Selon la classification internationale des brevets (CIB) ou à la fois selon la classification nationale et la CIB

B. DOMAINES SUR LESQUELS LA RECHERCHE A PORTE

Documentation minimale consultée (système de classification suivi des symboles de classement)
CIB 7 F24F

Documentation consultée autre que la documentation minimale dans la mesure où ces documents relèvent des domaines sur lesquels a porté la recherche

Base de données électronique consultée au cours de la recherche internationale (nom de la base de données, et si réalisable, termes de recherche utilisés)
EPO-Internal, WPI Data, PAJ

C. DOCUMENTS CONSIDERES COMME PERTINENTS

Catégorie °	Identification des documents cités, avec, le cas échéant, l'indication des passages pertinents	no. des revendications visées
A	US 3 243 890 A (K.H.EASTERDAY) 5 avril 1966 (1966-04-05) colonne 1, ligne 1 - ligne 9; figure 4	1,2,5, 7-12, 15-19
A	FR 2 753 780 A (REMLINGER FRANCOIS) 27 mars 1998 (1998-03-27) le document en entier	1
A	US 5 647 165 A (GUARGA FERRO RAFAEL) 15 juillet 1997 (1997-07-15)	

☐ Voir la suite du cadre C pour la fin de la liste des documents

☒ Les documents de familles de brevets sont indiqués en annexe

° Catégories spéciales de documents cités:

"A" document définissant l'état général de la technique, non considéré comme particulièrement pertinent
"E" document antérieur, mais publié à la date de dépôt international ou après cette date
"L" document pouvant jeter un doute sur une revendication de priorité ou cité pour déterminer la date de publication d'une autre citation ou pour une raison spéciale (telle qu'indiquée)
"O" document se référant à une divulgation orale, à un usage, à une exposition ou tous autres moyens
"P" document publié avant la date de dépôt international, mais postérieurement à la date de priorité revendiquée
"T" document ultérieur publié après la date de dépôt international ou la date de priorité et n'appartenant pas à l'état de la technique pertinent, mais cité pour comprendre le principe ou la théorie constituant la base de l'invention
"X" document particulièrement pertinent; l'invention revendiquée ne peut être considérée comme nouvelle ou comme impliquant une activité inventive par rapport au document considéré isolément
"Y" document particulièrement pertinent; l'invention revendiquée ne peut être considérée comme impliquant une activité inventive lorsque le document est associé à un ou plusieurs autres documents de même nature, cette combinaison étant évidente pour une personne du métier
"&" document qui fait partie de la même famille de brevets

Date à laquelle la recherche internationale a été effectivement achevée	Date d'expédition du présent rapport de recherche internationale
29 janvier 2001	05/02/2001

Nom et adresse postale de l'administration chargée de la recherche internationale	Fonctionnaire autorisé
Office Européen des Brevets, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31–70) 340–2040, Tx. 31 651 epo nl, Fax: (+31–70) 340–3016	Gonzalez-Granda, C

1

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
/BE 00/00122

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 3243890 A	05-04-1966	NONE	
FR 2753780 A	27-03-1998	NONE	
US 5647165 A	15-07-1997	ES 2063654 A	01-01-1995
		IT 1255171 B	20-10-1995

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
T1639-PCT		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/BE00/00120	06/10/2000	06/10/1999
International Patent Classification (IPC) or national classification and IPC C12N5/06		
Applicant TIGENIX N.V. et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 10 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 4 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations suporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
13/04/2001	03.01.2002
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Merckling, V Telephone No. +49 89 2399 8590

Form PCT/IPEA/409 (cover sheet) (January 1994)

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/BE00/00120

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:
Description, pages:

1-31	as originally filed	

Claims, No.:

1-30	with telefax of	08/10/2001

Drawings, sheets:

1-9	as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   - ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
   - ☐ the language of publication of the international application (under Rule 48.3(b)).
   - ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   - ☐ contained in the international application in written form.
   - ☐ filed together with the international application in computer readable form.
   - ☐ furnished subsequently to this Authority in written form.
   - ☐ furnished subsequently to this Authority in computer readable form.
   - ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
   - ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   - ☐ the description, pages:
   - ☐ the claims, Nos.:

☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application.

☒ claims Nos. 15-17,19-20,25,30.

because:

☒ the said international application, or the said claims Nos. 15-17,19-20,25,30 relate to the following subject matter which does not require an international preliminary examination (*specify*):
see separate sheet

☐ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. are so unclear that no meaningful opinion could be formed (*specify*):

☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

☐ no international search report has been established for the said claims Nos. .

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

☐ the written form has not been furnished or does not comply with the standard.

☐ the computer readable form has not been furnished or does not comply with the standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N) Yes: Claims 6,15,19,29

	No:	Claims	1-5,7-14,16-18,20-28,30
Inventive step (IS)	Yes:	Claims	6,19,29
	No:	Claims	1-5, 7-18,20-28,30
Industrial applicability (IA)	Yes:	Claims	1-14,18,21-24,26-29(YES),15-17,19-20,25,30 see separate sheet
	No:	Claims	

2. Citations and explanations
see separate sheet

1. Reference is made to the following documents :

   D1 : WO-A-9835022
   D2 : WO-A-9641620
   D3 : WO-A-9726326
   D4 : US-A-5811094
   D5 : WO-A-9607733
   D6 : US-A-5972703 (and corresponding document WO-A-9605290, published before the priority date of the present application and annexed to the communication)
   D7 : WO-A-9707200
   D8 : Zvi Nevo et al. (1998)
   D9 : WO-A-9928443
   D10 : Erlacher et al. (1998)
   Exhibits 1-5 : received 08.10.01 with the Applicant's letter of 08.10.01.

Regarding point V

2. **D1** is directed to a method for distinguishing undifferentiated human mesenchymal stem cells (hMSC) from partially or completely differentiated human mesenchymal cells comprising using $p21^{CIP1}$ as a marker for partial or complete differentiation, i.e. as a *negative marker* for selecting undifferentiated hMSC (see cl. 1 and page 16 last §)). Differentiated osteoblast precursors are also identified by screening with anti-ALK3 receptor antibodies, ALK3 receptor being used as a positive marker for differentiation (see ex. 1). The invention also provides hMSC in vivo implants combined with the administration of osteogenic induction agents (page 4), as well as methods for screening for cell inducers of cell differentiation such as BMPs (cl. 5-7). **D2** discloses methods for identifying mesenchymal skeletal progenitor cells comprising assaying for the presence of FGFR3 on said cells, i.e. using FGFR3 as a positive marker. Methods for isolating said cells, pure cell cultures and implants comprising said cells are also described (see cl. 1-11). The use of these cells (and/or implants containing them) for treating achondroplasia or joint surface defects is

mentioned (cl. 14 and page 7).

D3 describes a method for isolating precursor cells from a mammalian tissue comprising contacting them with an agent that binds the CD34 surface marker, and the use of said cells for promoting bone regeneration, either directly or in the form of an implant (see cl. 48 and 54).

In **D4**, the use of hMSC for producing connective tissue or treating a connective tissue defect is disclosed (cl. 1-2). The defect is preferably a cartilage or bone defect and the cells have preferably been culture-expanded (cl. 5-7 and 23). Prosthetic devices and implants containing these cells are also described (col. 4). Antibodies that specifically bind hMSC antigens are used for selecting and purifying hMSC (col. 2 line 57 to col. 3 line 3). Markers carried by differentiated mesenchymal phenotypes (chondrocytes, osteoblasts), such as type II collagen, BGP, Stro-1, are used as negative selection markers (see col. 38 line 53 to col. 39 line 34). It is also stated that hMSC express several growth factors (col. 44 line 66 to col. 45 line 32) and that dexamethasone induces the differentiation of hMSC into osteoblasts.

D5 solves the technical problem of providing certain immortalized stem cell precursors and/or fully differentiated cells that retain their differentiated phenotype while in culture. This problem is solved by preparing immortalized cells from a tissue of a growth suppressor gene (e.g. p53) deficient animal. In an embodiment, the tissue is bone marrow or calvarial bone and the isolated cells are osteoblast precursors or osteoclast precursors. These cells are selected with positive differentiation markers such as TRAP, calcitonin, collagen type IIB, albumin, osteocalcin etc... (see page 4, cl. 16-19 and page 5 lines 4-20). Such cells administered in vivo in a diffusion chamber induced bone mineralization (pages 37-38). however, osteocalcin andcalcitonin (etc...) are already markers of differentiated cells. No pluripotent cells were isolated by this method.

D6 describes a process for preparing an enriched population of bone precursor cells comprising exposing cells from the bone marrow to antibodies and retaining only the cells by to the antibodies. Said antibodies may be directed against several markers such as osteocalcin, osteonectin or bone alkaline phosphatase. CD34 is used as a negative selection marker (cl. 48). Preferably, the bone precursor cells are preosteoblasts (see cl. 48-50). Implants containing said selected cells and their use in bone repair is also disclosed (col. 11-12). Optionally, the implants are combined with bone growth factors promoting cell differentiation (col. 11 lines 32-55). These cells are not pluripotent, they were not shown to be capable of differenciating into

chondrocytes (see also comments on markers, **D5** above).
D7 describes a method for purifying adult oligodendrocyte precursors from a suspension derived from CNS comprising binding the cells to two different antibodies, the first one being directed to Thy-1 negative selection marker) and the second one being A2B5 antibody or NG-2 antibody (positive selection markers) (see cl.7).
D8 discloses implants of cultured cells for accelerating cartilage regeneration in defects of articular surfaces. Several types of cells were tested. Cells carrying the preskeletal/precartilaginous marker FGFR3 failed to induce a complete regeneration of the defects, whereas FGFR3-negative cells induced a complete regeneration (see abstract). Typical markers of precartilaginous cells, useful for their selection/enrichment, are FGFR3 at the cell surface and collagen type IIA in the extracellular matrix (ECM) (see page 63 right col.).
D9 describes a method of maintaining a cartilaginous phenotype in chondrocytes in vitro comprising culturing said chondrocytes in a serum-free medium comprising a cartilage-derived morphogenetic protein such as a BMP, CDMP-1 or CDMP-2 (see cl. 26). These chondrocytes ae used for repairing joint surface defects in mammals (cl. 27).

2.1 As far as can be understood, claim 1 is directed to a screening method for identifying precursor cells by positively identifying a marker protein. Since the term "morphogenic protein" (broader than "bone morphogenic protein" or "cartilage-derived morphogenic protein", see exhibit 2) is not clear, it is not possible to determine unambiguously which proteins are covered by this expression. Claim 2 is directed to the same subject-matter, except that the screening method uses a negative marker, i.e. the absence of a certain marker, for identifying the precursor cells.

2.2 D2 more precisely discloses the detection of FGF-R3 positive bone tissue in vivo (with anti-FGF-R3 antibodies) and the use of said tissue for obtaining in vitro cell culture (see examples 1-3). These cells, identified as mesenchymal progenitor cells, grow more rapidly than cells obtained from FGF-R3 negative tissues, and their use for making implants and repairing bone defect is claimed. On the other hand, Sasse et al. (exhibit 3) shows that chondrocytes from fetal, newborn or adult bovine cartilage all synthesize FGF-R3 RNA in vitro when cultured at high density. The

FGF-R3 RNA level becomes undetectable at low cell density. it is concluded that FGF-R3 represents a new marker for following chondrocyte maturation and differentiation. These results nevertheless are based on chondrocyte cultures that were obtained by an undescribed method and that may contain a mixture of cells at various stages of differentiation. The lack of clarity of the experimental procedure and of the conclusions drawn from the results might be a reason why this document (which is a poster abstract) was never published as a reviewed paper (see arguments found in exhibit 4, especially points 1.7, 1.9, 1.10 and 1.12). In any case, the Sasse abstract being the only document with results that may contradict D2, it does not seem sufficient to teach away from D2. Finally, it should be stressed that FGF-R3 is not excluded from the scope of present claim 1. Should the use of FGF-R3 as a positive selection marker be regarded as ineffective for the purpose of the application, it should not fall within the scope of the claims.

As for D3, even if CD34 is also expressed on hematopoietic stem cells, the object of D3 is to show that this marker can be used for positive selection of bone and cartilage precursor cells (even if other cells may also be isolated by this method) (see page 8 lines 8-11 and page 12 line 19 to page 14 line 11). The "degree of efficiency", i.e. number of passages after which the marker remains expressed on the cells, of the selection method is irrelevant for assessing novelty. Should the Applicant consider that CD34 cannot be used at all as a positive marker for the purpose of the invention, he should be reminded that CD34 is not excluded from the scope of claim 1 of the present application.

Concerning D4, this document provides some explanations for the absence of chondrogenesis (see col. 25 lines 16-30) and also shows that the cells obtained with the method of the invention are not differentiated chondrocytes or osteocytes (see col. 38 line 38 to col. 39 line 34, and table 5, to be compared to figure 2 of exhibit 1). There is no reason to believe that these cells are not bone marrow-derived mesenchymal stem cells. Although the selection markers themselves are not fully defined, the disclosure of D4 is enabling because the monoclonal antibodies are well defined (deposit of hybridomas, see col. 25 lines 43-50).

D2-D4 are regarded as disclosing a method as defined in claim 1. The positive markers used are : FGFR3 in D2, CD34 in D3 and defined antibodies in D4. As for claim 2, it is anticipated by D2 and D4. These documents use the following negative selection markers : type II collagen in D4, $p21^{CIP1}$ in D2, FGF-R3 in D8.

All of D2-D4relate to skeletal precursor cells, and the selection marker is a protein in all documents, so that claims 4-5 are not novel either. Claim 3 is anticipated by D8 (see above).
For the same reasons, claims 7, 21-22, 25 and 26-28,30 are not new either.

2.2 All of the screening/enriching methods described inD2-D4 use antibodies recognizing cell surface markers, the subject-matter of claims 8-11 is not new.

2.3 It should be stressed that, in claim 12, the precursor cells are defined as "marked according to claims 1-7". However, cells cannot be rendered novel by their isolation process, so that the use of precursor cells that have been isolated/purified using another method for repairing connective tissue in mammals is also novelty-destroying for claim 12. Such a use is thus anticipated by D2-D4. Claims 12-14 are not new.
The latter documents also describe the use of precursor cells in the form of implants or prosthetic devices, in combination with a matrix, for use for treating for example a traumatic lesion in bone or cartilage. Claims 16-18 are not new. For the same reason, claims 20 and 23-24 are not new either.

2.4 None of the available documents discloses a screening method using CDMP-1 or a TGF-β having 80% homology with CDMP-1 as a positive marker. Claims 6 and 29 are novel.
The prior art does not explicitly disclose the use of precursor cells as a source of growth factor so that claim 15 is new.
The prior art does not anticipate claim 19 because no method for repairing joint surface defects comprising the co-implantation of skeletal precursor cells and chondrocytes is disclosed.

3. Regarding claims 6 and 29, D10 would be the closest prior art. It discloses that CDMP-1 and CDMP-2 induce de novo cartilage and bone formation in vivo and states that CDMP-2's high levels of expression in postnatal cartilaginous cells suggest a possible role in the promotion and maintenance of the cartilaginous phenotype (abstract and page 383 right col. to page 384 left col.). The person skilled in the art attempting to solve the problem of providing a screening method for identifying precursor cells would at the most regard CDMP-2 as a potential candidate

for a precursor cell marker. However, there are no precise data, the statement only relates to postnatal cartilaginous cells does not mention CDMP-1. The subject-matter of claims 6 and 29 would therefore not be obvious over D10.

3.1 D4 does not explicitly disclose the use of precursor cells as a "source" of growth factors but clearly discloses that such cells express growth factors. The subject-matter of claim 15 appears to be obvious over the teaching of D4 for the person skilled in the art.

3.2 None of the available documents teaches or suggests to implant a combination of precursor cells and differentiated chondrocytes in a mammal for repairing a joint surface defect. Claim 19 would appear to be inventive.

4. For the assessment of the present claims 15-17, 19-20 and 25, 30 on the question whether they are industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can also be dependent upon the formulation of the claims. The EPO, for example, does not recognize as industrially applicable the subject-matter of claims to the use of a compound in medical treatment, but may allow, however, claims to a known compound for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment.

Regarding point III

5. Claims 15-17, 19-20 and 25, 30 relate to subject-matter considered by this Authority to be covered by the provisions of Rule 67.1(iv) PCT. Consequently, no opinion will be formulated with respect to the industrial applicability of the subject-matter of these claims (Article 34(4)(a)(i) PCT).

CLAIMS

1. Method of positively identifying viable, committed, pluripotent skeletal precursor cells that have entered a post-natal differentiation pathway leading to skeletal or connective tissues comprising the steps of:

   isolating mammalian cells into a cell culture in vitro, and

   detecting the presence of a positive embryonic marker of an expressed morphogenic protein, a homolog thereof or a marker co-expressed and/or co-detectable with this marker.

2. The method according to claim 1, wherein the presence of the positive marker is further characterised by the absence of a negative marker.

3. The method according to claim 2, wherein the negative marker is FGFR3 or a marker or factor co-expressed or co-detectable with this negative marker.

4. The method according to any previous claim wherein the positive marker is an actively expressing gene, a protein or an mRNA expressed by a gene in the precursor cells or a part thereof, detectable at the DNA, mRNA, cDNA or the protein level and/or detectable via the activity of a promoter directing/regulating this gene expression, operably linked to a heterologous reporter gene.

5. The method according to any previous claim wherein the positive marker identifies precursor cells belonging to a joint interzone in mammals.

6. The method according to any previous claim wherein the expressed morphogenic protein is the cartilage-derived morphogenic protein CDMP-1 or a transforming growth factor β having at least 80% homology with CDMP-1 as a marker of skeletal precursor cells from any part of the body or a marker or factor co-expressed or co-detectable with any or all of these positive markers.

7. A method according to any previous claim, wherein
the step of detecting the presence of the positive marker includes applying a

binding agent for the positive marker to an isolated source of cells having the precursor cells, the marker positively identifying the viable skeletal precursor cells; and

separating the cells which are bound to the binding agent.

8. Use of reagents, ligands, and/or antibodies recognizing cell surface markers for sorting and enriching of precursor cells in cell culture in vitro, wherein the cell surface marker is co-expressed or co-detectable with the marker of any of claims 1 to 7.

9. Use of reagents, ligands, and/or antibodies recognizing cell surface markers according to claim 8, for sorting of skeletal precursor cells and for enriching a population in skeletal precursor cells.

10. Use for enriching via cell sorting according to claim 8 or 9, wherein cell sorting methods comprise fluorescence activated cell sorting, the use of magnetic beads coated with the respective antibodies or ligands, the use of affinity chromatography or the use of any other means coated with antibodies or ligands directed to the cells which are to be selected.

11. Use of reagents and/or antibodies according to any of claims 8 to 10 wherein the antibodies are polyclonal or monoclonal antibodies.

12. Use of skeletal precursor cells marked according to any of claims 1 to 7 for producing or repairing connective tissue in a mammal.

13. Use according to claim 12, wherein the said cells are cultured at a cell density of at least 10^5 cells/ml.

14. Use according to claim 12 or claim 13, comprising further administration of a factor that stimulates differentiation of the skeletal precursor cells into the type of connective tissue to be produced or repaired.

15. Use of precursor cells marked according to any of claims 1 to 7 as a source of

growth factors.

16. Use of precursor cells marked according to any of claims 1 to 7 as matrix producing cells.

17. Use according to claim 16, wherein the said matrix further comprises a bio-resorbable polymer or carrier.

18. Use according to claim 16 or 17 for the treatment of subglottic stenosis, tracheomalacia, chondromalacia patellae, osteoarthritis and traumatic lesions in a mammal.

19. A procedure for joint surface defect repair in a mammal comprising the co-implantation of skeletal precursor cells marked according to any of claims 1 to 7 and chondrocytes.

20. A method for enhancing the implantation of a prosthetic device in connective tissue comprising the step of implanting a prosthetic device having skeletal precursor cells according to any of the claims 1 to 7 adhered thereto under conditions suitable for differentiating the cells into the connective tissue desired.

21. A culture of isolated and expanded, viable, differentiated, pluripotent, precursor cells that have entered a post-natal differentiation pathway leading to skeletal or connective tissue, wherein the cells express a positive embryonic marker which is an expressed morphogenic protein, a homolog thereof or a marker co-expressed and/or co-detectable with this marker.

22. A therapeutic composition comprising the cells of claim 21.

23. An implant comprising the cells of claim 21.

24. The implant of claim 23 suitable for connective tissue implantation.

25. A method of treating a patient in need thereof comprising administration of the

therapeutic composition of claim 22.

26. A diagnostic for positively identifying in vitro a positive marker of viable, committed, pluripotent, skeletal precursor cells that have entered a post-natal differentiation pathway leading to skeletal or connective tissues, wherein the marker is an expressed morphogenic protein, a homolog thereof or a marker co-expressed and/or co-detectable with this marker.

27. The diagnostic according to claim 26 wherein the diagnostic also indentifies the absence of a negative marker.

28. The diagnostic wherein the positive marker identifies precursor cells belonging to a joint interzone in mammals.

29. The diagnostic according to any of claims 26 to 28 wherein the expressed morphogenic protein is the cartilage-derived morphogenic protein CDMP-1 or a transforming growth factor β having at least 80% homology with CDMP-1 as a marker of skeletal precursor cells from any part of the body or a marker or factor co-expressed or co-detectable with any or all of these positive markers.

30. Use of an embryonic marker to positively identify viable, committed skeletal pluripotent precursor cells that have entered a post-natal differentiation pathway leading to connective or skeletal tissues, wherein the embryonic marker is an expressed morphogenic protein, a homolog thereof or a marker co-expressed and/or co-detectable with this marker.